

Enzymatic Cleanup of PCR Products

1. Remove excess primers and dNTPs by digesting them with Exonuclease I (Exo), and Shrimp Alkaline Phosphatase (SAP). Aliquot and mix according to the following:

	Per 10ul sample	Per plate	Per 20 plates	Per 35ul sample	Per plate 35ul spl	Per 20 plates, 35ul spls
SAP (1Unit/ul)*1	0.37	37.00	740.00	1.30	129.50	2590.00
Exo I (10Units/ul)*2	0.19	19.00	380.00	0.67	66.50	1330.00
dH2O	1.94	194.00	3880.00	6.79	679.00	13580.00
Total	2.50	250.00	5000.00	8.75	875.00	17500.00
Aliquot per well	2.50	2.50	2.50	8.75	8.75	8.75
Aliquot per well on robot plate	-	-	52.50	-	-	175.00

2. Incubate at 37° C for 15 minutes for digestion of primers and dNTPs, then at 72° C for 15 minutes to kill the enzymes.

The PCR product is now ready for the usual sequencing with dye primers or dye terminators.

*1: SAP from Amersham, CATALOG # E 70092X

*2: ExoI from Amersham, CATALOG # E 70073Z

Cost is approximately \$25.00/96-well plate